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(54) Title: STABLE RECOMBINANT YEASTS FOR FERMENTING XYLOSE TO ETHANOL

(57) Abstract

Described are recombinant yeast which ferment xylose to ethanol and which maintain their ability to do so when cultured for numerous generations in non-selective media. The preferred yeast contain multiple copies of integrated genes encoding xylose reductase, xylitol dehydrogenase, and xylulokinase fused to promoters which are non-glucose inhibited and which do not require xylose for induction. Also described are preferred methods for integrating multiple copies of exogenous DNA into host cells by transforming cells with replicative/integrative vectors, and then replicating the cells a number of times under selective pressure to promote retention of the vector in subsequent generations. The replicated vectors thus serve to integrate multiple copies of the exogenous DNA into the host cells throughout the replication/selection phase. Thereafter the selective pressure can be removed to promote loss of the vector in subsequent generations, leaving stable integrants of the exogenous DNA.